(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
14 October 2004 (14.10.2004)

PCT

(10) International Publication Number
WO 2004/087877 A3

(51) International Patent Classification[7]: C12Q 1/68, A01N 43/04, C07H 21/04, A61K 31/07

(21) International Application Number: PCT/US2004/009261

(22) International Filing Date: 25 March 2004 (25.03.2004)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
60/457,745 26 March 2003 (26.03.2003) US
60/497,784 26 August 2003 (26.08.2003) US

(71) Applicant *(for all designated States except US)*: **ASTRAL INC.** [US/US]; 6175 Lusk Blvd., San Diego, CA 92121 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **SMITH, Dan** [US/US]; 4004 Caminito Meliado, San Diego, CA 92122 (US). **BOT, Adrian** [RO/US]; 27003 Carmelita Dr., Valencia, CA 92121 (US).

(74) Agent: **WURST, John, E.**; Astral Inc., 6175 Lusk Blvd., San Diego, CA 92128 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*
— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

(88) Date of publication of the international search report: 16 June 2005

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: SELECTED RNA MOTIFS TO INCLUDE CELL DEATH AND/OR APOPTOSIS

(57) Abstract: The present application is directed to the use of dsRNA and/or ssRNA for the purpose of inducing apoptosis or cell death in proliferating cells. Specifically, low molecular weight and high molecular weight dsRNA and ssRNA are shown to induce apoptosis and/or cell death in proliferating cells, to arrest proliferation of transformed cells or tumor cells and to cause rapid induction of the cytokine TNF-alpha and/or also induce production of IL-12 which directs a Th-1 response.

WO 2004/087877 A3

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/09261

A. CLASSIFICATION OF SUBJECT MATTER
IPC(7) : C12Q 1/68; A01N 43/04; C07H 21/04; A61K 31/07
US CL : 435/6, 91.1, 325, 375; 536/24.3, 24.33, 24.5; 514/44
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 435/6, 91.1, 325, 375; 536/24.3, 24.33, 24.5; 514/44

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	ELBASHIR et al. RNA interference is mediated by 21- and 22-nucleotide RNAs. Genes and Development, 2000 Vol. 15:188-200, see RNA synthesis and cloning of 21 nucleotide RNAs, at page 198.	28-37
X --- A	WIANNY et al. Specific interference with gene function by double-stranded RNA in early mouse development. Nature Cell Biology, 2000 Vol. 2:70-75, see RNA synthesis at page 74.	28-37 --- 1-27
X	SVOBODA et al. Selective reduction of dormant maternal mRNAs in mouse oocytes by RNA interference. Development, 2000 Vol. 127:4147-4156, see dsRNA preparation at page 4148.	28-37
X	ZELEZNICK et al. Treatment of leukemic (L-1210) Mice with Double-stranded Polyribonucleotides. Proc Soc Exp Biol Med, 1969, Vol 130:126-128.	1-37
A	BRANCH, AD. A good antisense molecule is hard to find. TIBS, 1998 Vol. 23:45-50, see entire article.	1-27
A	JEN et al. Suppression of gene expression by targeted disruption of messenger RNA: Available options and current strategies. Stem Cells, 2000 Vol. 18:307-319, see entire article.	1-27

☒ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"B" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search
13 March 2005 (13.03.2005)

Date of mailing of the international search report
27 APR 2005

Name and mailing address of the ISA/US
Mail Stop PCT, Attn: ISA/US
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450
Facsimile No. (703) 305-3230

Authorized officer
Terra C. Gibbs

Telephone No. 571-272-0564

Form PCT/ISA/210 (second sheet) (January 2004)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/09261

C. (Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	OATES et al. Too much interference: Injection of double-stranded RAN has nonspecific effects in the zebrafish embryo. Developmental Biology, 2000 Vol. 224:20-28.	1-27
A	COBURN et al. siRNAs: a new wave of RNA-based therapeutics: Journal of Antimicrobial Chemotherapy, 2003 Vol. 51:753-756.	1-27
A	AGAMI, A. RNAi and related mechanisms and their potential use for therapy. Current Opinion in Chemical Biology, 2002 Vol. 6:829-834.	1-27
A	GREEN et al. Antisense Oligonucleotides: An evolving technology for the modulation of gene expression in human diseases, 2000. J Am Coll Surg Vol. 191:93-105.	1-27
A	AGRAWAL et al. Antisense therapeutics: is it as simple as complementary base recognition? Molecular Medicine Today, 2000 Vol. 61:72-80.	1-27

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/09261

Continuation of B. FIELDS SEARCHED Item 3:
CaPlus, EmBase, CancerLit, Medline, NPL, WEST
search terms: double-stranded RNA (dsRNA), single-stranded RNA (ssRNA), short interfering RNA (siRNA)